Introduction: In cancer patients, coadministration of methotrexate (MTX) and proton pump inhibitors (PPIs) can cause a pharmacokinetic interaction for delayed elimination and a subsequent increase in blood MTX concentration. Human organic anion transporters hOAT1 (*SLC22A6*) and hOAT3 (*SLC22A8*) are responsible for renal tubular secretion of MTX. We hypothesized that they are involved in drug interaction of MTX with PPIs. The aim of this study was to evaluate the inhibitory potencies of PPIs on HEK-hOAT1 and HEK-hOAT3 attempt to give an explanation to this possible drug–drug interaction between MTX and PPIs.
Patients (or Materials) and Methods: Uptake experiments were performed using HEK Flp-In 293, stably expressing human OAT1*(SLC22A6)* or OAT3 *(SLC22A8)*, by using the Flp-In™ System recombinase (Invitrogen®). We analyzed whether the inhibitory potencies of omeprazole, lansoprazole, and pantoprazole on OAT-mediated [3H] MTX uptake in vitro.
Results: MTX shown to be a high-affinity substrate for hOAT3 but not for hOAT1 (hOAT3 Km = 21.17 [5.65] μM). All PPIs showed in vitro inhibition of hOAT transporters. For that purpose, PAH, and ES were selected for probing hOAT1 and hOAT3 activities. However, their IC50 values concerning hOAT1 were 10 to 30 times higher than the unbound plasma concentrations of the PPIs. Conducted in parallel [3H]MTX uptake into HEK hOAT3 cells was inhibited by all PPIs in a concentration-dependent manner omeprazole, lansoprazole, and pantoprazole inhibit hOAT3, their IC50 were in the range of the therapeutic levels of the IPPs (0.91–8.06 μM).
Conclusion: PPIs significantly affect transport of MTX mediated by hOAT3, but this interaction cannot be explained by the inhibitory effects of PPIs on renal hOAT1. These in vitro results demonstrate that alterations of uptake transporters of MTX function by IPPs drugs have to be considered as potential mechanisms underlying drug–drug interactions between MTX-IPPs.
Disclosure of Interest: None declared.

PP183—EFFECT OF RITONAVIR, KETOCONAZOLE AND RIFAMPIN ON INTESTINAL AND HEPATIC CYTOCHROME P450 3A ENZYMES IN HEALTHY ASIAN ADULTS

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Introduction: Cytochrome P450 3A (CYP3A) is the major enzyme metabolizing drugs and xenobiotics in humans. The present study aimed to quantitate the extent of in vivo inhibition and induction effects of ritonavir, ketoconazole, and rifampin on the intestinal and hepatic activity of CYP3A in adult healthy Asians.
Patients (or Materials) and Methods: Fifteen healthy male or female Asian adults completed this open-label, single-center, sequential, partial crossover study. Subjects underwent 4 periods of treatments: baseline (reference) period; period 1, either ketoconazole 200 mg or ritonavir 100 mg twice daily given for 3 days; period 2, either ritonavir or ketoconazole given for 3 days (periods 1 and 2 in a randomized crossover design); period 3, rifampin 600 mg given nightly for 2 weeks. At the end of each period, each subject was administered with a single intravenous (IV) midazolam 0.75 mg, given in the fasting state, and a single oral 1.5-mg dose of midazolam 4 hours later. Serial blood samples were taken from the subjects at predose and at various time points for up to 12 hours after IV administration of midazolam. Concentrations of midazolam were measured using a validated LC/MS-MS approach. The hepatic midazolam clearance after IV dosing was directly modeled in NONMEM and was estimated separately for the reference and the test periods. The intestinal midazolam availability was calculated by dividing oral midazolam availability with hepatic midazolam availability. Formal interaction was excluded if the 90% CI for the ritonavir, ketoconazole, or rifampin over reference ratios for phenotyping metrics (hepatic midazolam clearance and intestinal midazolam availability) was within a 0.80 to 1.25 range.
Results: Ritonavir reduced hepatic and intestinal CYP3A activity to 0.68-fold (90% CI, 0.61–0.73) and to 0.36-fold (0.28–0.52), respectively. Ketoconazole reduced hepatic and intestinal CYP3A activity to 0.75-fold (90% CI, 0.69–0.81) and 0.52-fold (0.39–0.83), respectively. Rifampin increased hepatic and intestinal CYP3A activity by 1.22-fold (90% CI, 1.02–1.42) and 1.41-fold (1.04–2.19), respectively. There was a statistically significant and clinically relevant inhibition of the intestinal CYP3A activity due to chronic treatment with ritonavir.
Conclusion: Ritonavir treatment resulted in a clinically significant reduction in the net intestinal CYP3A activity. Extent of CYP3A inhibition produced by ketoconazole was smaller relative to ritonavir, especially at the intestinal level.
Disclosure of Interest: None declared.

PP186—THE ANALYSIS OF INTERACTION OF WARFARIN IN THE REAL CLINICAL PRACTICE

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Introduction: Estimation of prescription of warfarin (W) taking into account its interaction with other medicines in patients with coronary heart disease in clinical conditions.
Patients (or Materials) and Methods: The design of the research is retrospective. To estimate the level of polypharmacy and to analyze the interaction of W with other medicines medical documents of 61 patients with coronary heart disease have been analyzed. The effects of interaction of medicines were estimated according to BNF (British National Formulary) in March 2013. Estimation of the level of polypharmacy was considered according to the following 3 groups: 1, those who received 1 to 5 medicines. This shows rational use of medicines. 2, those who received 6 to 10 medicines; 3, those who received 11 to 15 medicines. The most clinically significant combinations of medicines have been analyzed.
Results: It has been established that in total 61 patients received 709 medicines. The medicine load for 1 patient was 11.6 medicines; 77% of the patients received from 11 to 15 medicines, 23% of the patients received from 6 to 10 medicines. The research did not reveal any patients who received ≤5 medicines. The results testified to a high level of polypharmacy. The next stage is the study of combinations of W and medicines presenting significant risk in terms of clinical safety. W was given in combination with the following medicines: heparin – 16% of patients; acetylsalicylic acid (AS acid) – 28% of patients; omeprazole – 21%; diclofenac – 13%; ciprofloxacin – 11%; metronidazole – 5%. It has been proved that the combinations of W and heparin, AS acid, and omeprazole are inadmissible, because they are life-threatening due to hemorrhage complications. Other unfavorable combinations of W have been revealed. They are: W + AS acid + omeprazole – 10% of patients; W + AS acid + ciprofloxacin – 4%; W + AS acid + diclofenac – 14%. These combinations significantly increase the risk of gastrointestinal bleeding.
Conclusion: The real clinical practice in hospitals revealed irrational prescription of medicines. The research has registered a high level of polypharmacy and dangerous combinations of W with other medicines. It has been proved that before prescribing treatment with W it is necessary to consider pharmacology genetics options, which are widely used worldwide. This kind of research has not been done in Kyrgyzstan. This determines the importance of personalized approach to pharmacologic therapy of W. This

work is the first stage for determining the design of pharmacology genetics research.

Disclosure of Interest: None declared.

PP189—A PHYSIOLOGICALLY-BASED MECHANISTIC PHARMACOKINETIC MODEL TO ASSESS THE METABOLISM OF OXYCODONE IN HEALTHY VOLUNTEERS: INTERPLAY BETWEEN CYP3A AND 2D6 INHIBITION

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Introduction: Oxycodone undergoes a relatively complicated metabolism producing 3 main metabolites: noroxycodone, oxymorphone, and noroxymorphone. Among these metabolites, oxymorphone is highly related to the pharmacodynamic effect of oxycodone. It is 14 times more potent than the parent compound, and its affinity for the μ opioid receptor is 3 times higher than morphine. Development of a whole-body physiologically based pharmacokinetic (PK) model is an approach to predict in vivo metabolism of oxycodone and PK profile of each metabolite in different drug–drug interaction (DDI) scenarios.

Patients (or Materials) and Methods: The Simcyp simulator was used as a platform and database for simulation of oxycodone's metabolism in virtual healthy populations. Prior in vitro and in vivo data were combined to build an oxycodone model which was used to predict the PK profile in healthy volunteers. The incorporated parameters were optimized by a top-down approach based on the clinical trial conducted by Samer et al, where PK profile of 0.2 mg/kg single dose oxycodone and its 3 metabolites were monitored in 10 healthy male volunteers (n = 10) previously genotyped for CYP2D6 in 4 scenarios (oxycodone administered alone or coadministered with CYP3A inhibitor ketoconazole (400 mg) and/or CYP 2D6 inhibitor quinidine (100 mg)) (Samer C, Daali Y et al. 2010). The PK data obtained in 3 interaction scenarios of the latter clinical trial were used to test the built model. Simulated trials permitted to evaluate the impact of CYP3A and CYP2D6 inhibitions on the concentration–time profiles of oxycodone and 3 main metabolites. The simulated studies designs were closely matched with the clinical trial and the virtual populations (1 trial of 10 volunteers, and 10 trials of 10 volunteers) were set with the same proportion of each CYP2D6 phenotype as the clinical trial (7 extensive, 1 poor and 2 ultrarapid metabolisers).

	AUC_{24h} (ng·min/mL)			C_{max} (ng/mL)		
	Observed	Simulated	Simulated	Observed	Simulated	Simulated
	n = 10	n = 10	n = 100	n = 10	n = 10	n = 100
Oxycodone	12,086 (3184)	12,305 (8002)	12,953 (6088)	35.9 (11.3)	32.2 (14.8)	35.3 (13.6)
Noroxycodone	7753 (2127)	7101 (4442)	7502 (4875)	15.5 (4.3)	14.3 (6.7)	15.3 (9.7)
Oxymorphone	232 (142)	179 (145)	244 (267)	0.7 (0.4)	0.7 (0.7)	0.8 (0.7)

Results: Pharmacokinetic profiles of oxycodone and 2 predominant metabolites (oxymorphone and noroxycodone) were closely simulated by the model.

Mean values (SD).

Oxycodone, noroxycodone, and oxymorphone PK profiles were also concordant with the clinical study according to CYP2D6 phenotypic groups. Obtained DDI magnitudes were also in agreement with the clinical data. Noroxymorphone PK profile was less accurately predicted by the model.

Conclusion: The Simyp developped model for oxycodone is valuable to predict the metabolism of oxycodone and main metabolites, and to simulate DDI involving CYP 3A and 2D6.

Disclosure of Interest: None declared.

PP191—INFLUENCE OF VERAPAMIL ON THE PHARMACOKINETICS OF OXCARBAZEPINE AND 10-HYDROXYCARBAZEPINE ENANTIOMERS IN HEALTHY VOLUNTEERS

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Introduction: Oxcarbazepine (OXC) is a drug indicated for the treatment of partial seizures or generalized tonic-clonic seizures in adults and children. It undergoes rapid presystemic reduction with formation of the active metabolite 10-hydroxycarbazepine (MHD), which has a chiral center at position 10, with the enantiomers (S)-(+)- and R-(-)-MHD with similar antiepileptic effects. OXC and MHD are substrates of P-glycoprotein (Pgp), whereas verapamil is an inhibitor of Pgp expressed in various tissues, including the brain. This study aims to evaluate the influence of verapamil on the pharmacokinetics of OXC and MHD enantiomers in healthy volunteers.

Patients (or Materials) and Methods: The study was conducted in 2 phases and included 12 adult healthy volunteers. In the Phase I, they were treated with 300 mg/12 hours OXC during 5 days. On the fifth day, after the last dose, serial blood samples were collected up to 12 hours. In the Phase II, the same healthy volunteers were treated with OXC (300 mg/12 hours during 5 days) associated with verapamil (80 mg/8 hours during 5 days). On the fifth day, after the last OXC dose, serial blood samples were collected up to 12 hours. Plasma concentrations of OXC and MHD enantiomers were evaluated by LC-MS/MS coupled with a chiral phase Chiralcel® OD-H column. Pharmacokinetic analysis was performed using the software WinNonlin and statistical tests were conducted with the significance level set at $P < 0.05$.

Results: The following pharmacokinetic parameters for OXC were obtained in Phase I (median): maximum plasma concentration (Cmax) of 1.35 mg/mL in 1.0 hour, area under the plasma concentration versus time curve (AUC0–12) of 3.98 μg·h/mL and half-life of 2.45 hours. The kinetic disposition of MHD was enantioselective, with observation of a higher proportion for the enantiomer S-(+)-MHD compared with R-(-)-MHD (AUC0-12S-(+) / R-(-) of 4.10). Verapamil treatment (Phase II) decreased the mean residence time (3.83 vs 4.71 hours) and the apparent volume of distribution (Vdárea /f')(2.86 vs 3.78 L/kg) of OXC. Concerning MHD enantiomers, the verapamil treatment increased Cmax, AUC and Css for both enantiomers.

Conclusion: Verapamil treatment reduced OXC Vdárea/f' and increased AUC of both MHD enantiomers probably due to the Pgp inhibition.

Disclosure of Interest: None declared.